

THE REACTION OF TWO STARFISHES, *PATIRIA MINIATA* AND *ASTERIAS FORBESI*, TO FOREIGN TISSUE IN THE COELOM¹

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The reactions of invertebrate animals to invasion of the body by foreign materials are not clearly understood, largely because relatively little attention has been focused on this aspect of invertebrate biology. Phagocytosis appears to be the most common defense (Cantacuzène, 1923; Huff, 1940) and has been described by many workers, among them Durham (1888, 1891), Metchnikoff (1891), Kindred (1921), Lison (1930), and Boolootian and Giese (1958). These authors and others note that large objects, such as clumps of carmine which cannot be ingested by individual phagocytes, are generally encapsulated or surrounded by layers of amebocytes. Very little direct work has been done on the specific reactions of invertebrates to large implants, however; the results of Labbe (1929) working on molluscs, Cameron (1932) working on earthworms, Salt (1957) working on insects, and Triplett, Cushing and Durall (1958) working on sipunculid worms, are perhaps typical and suggest that encapsulation may be a general invertebrate reaction toward all foreign objects too large to be phagocytized.

Triplett, Cushing and Durall note that the sipunculid worm, *Dendrostomum zostericum*, discriminates between pieces of sipunculid and sea anemone tentacles introduced into the coelom, for although both are encapsulated, the sea anemone tissue is killed whereas the sipunculid tentacles are recovered from their capsules alive. The worms are evidently incapable, however, of distinguishing between pieces of sipunculid tentacle of homologous and heterologous origin. The data of Cushing (1957), who made autologous and homologous grafts of eyes to the female portions of the gonads in the scallop, *Aequipecten irradians*, suggest that the hosts are slightly more tolerant of autologous than of homologous grafts, but Cushing points out that his results are preliminary and need extension and confirmation. Anderson (unpublished) performed a brief series of exploratory studies in which he introduced pieces of pyloric caecum from the starfishes, *Pisaster ochraceus* (Order Forcipulata) and *Henricia leviuscula* (Order Spinulosa) into the coelom of another starfish, *Patiria miniata* (Order Spinulosa). He was able to recover the *Pisaster* caecum in apparently healthy condition after ten days in the foreign environment. The *Henricia* caecum, on the other hand, was found to have undergone almost complete disintegration. These results are only preliminary, but they

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present a contrast to the general pattern of encapsulation described above. They also serve to illustrate the inconsistency of reactions in invertebrate groups, for the *Pisaster* tissue, which was tolerated, is less closely related to the host (in a different order) than the *Henricia* caecum, which was rejected.

The present study was initiated as a further preliminary exploration of the nature of the reactions of starfishes to foreign tissue in the coelom, in the hope of determining whether the host distinguishes between the tissues of donors of different degrees of relationship, and how the foreign tissues are treated. For many reasons, the asteroids serve as excellent experimental material in transplant studies. The coelom is large and easily reached by knife and hypodermic needle. The coelomic fluid is similar to sea water in composition and tonicity, and transplants may be handled or held in ordinary sea water for considerable periods of time without being damaged. For the same reason, leakage of sea water through the implantation incision in the host body wall does not greatly alter the internal environment of the animal. The animals are generally hardy and regenerate easily. They can withstand the loss of pieces of body wall or of viscera, and they heal experimental incisions well. The pyloric caeca, which are suspended in the coelom by mesenteries, provide excellent tissue for transfer. Caecum tissue is relatively active metabolically and shows a high rate of molecular turnover (Ferguson, 1964a, 1964b) so that its presence has a considerable effect on the composition of the coelomic fluid. It is easily removed from the donor. Above all, it is quite distinctive histologically; in addition to classical accounts, more recent descriptions of normal histology have been provided for *Asterias forbesi* (Anderson, 1953) and *Henricia leviuscula* (Anderson, 1960, 1962). Hence, it is possible to determine by histological study whether a sample seems to be normal and functioning, or to detect changes that may have taken place as a result of sojourn in a host animal.

The general questions toward which this study was directed were the following:

- (1) Does a host starfish distinguish between implanted pieces of pyloric caecum from donors of different degrees of relationship?
- (2) What is the nature of the animal's reaction to foreign tissue, and by what means (if any) are large pieces removed from the coelom?
- (3) What effect, if any, does sojourn in a strange environment have upon tissue of different donors?

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MATERIALS AND METHODS

The problem involved introducing pieces of pyloric caecum of one animal (donor) into the coelom of another (host) and determining the fate of such transplants. The host species used were *Patiria miniata*, obtained from the central California coast, and *Asterias forbesi*, obtained from Woods Hole, Massachusetts. Fairly small specimens, ranging from one to three inches in diameter, were used because they required relatively little aquarium space and their body walls were

thin and easy to cut. Donors included members of the host species and of two others, *Asterias vulgaris* and *Henricia sanguinolenta*, both obtained from Woods Hole. The animals were kept in running sea water at the Marine Biological Laboratory at Woods Hole, where most of the experiments on *Asterias* were done, and in tanks of circulating refrigerated sea water at Cornell University, where the balance of the experiments on *Asterias* and all those on *Patiria* were performed.

Before being subjected to any experimental treatment, both hosts and donors were first relaxed in a solution of 7.5–8% MgCl_2 in tap water. Pyloric caecum was then removed from the donors and, in order to distinguish it from the caecum of the host, was stained for at least two minutes in 0.01% methylene blue in sea water, to which had been added 0.03% egg albumin to reduce the toxicity of the dye (Chalkley and Park, 1947). The fact that homologous tissues stained in this way were not rejected by the host indicates that this treatment does not significantly alter the chemistry of the transplants. Implanted tissues retained the blue color for more than six weeks and thus were easy to identify in gross dissection; the stain was completely removed, however, by subsequent histological procedures. The staining step was omitted when *Henricia* was the donor, for the caecum of this species is brilliant red-orange in color and is easy to identify without any special treatment.

The tissue was trimmed and injected by means of a large-bore hypodermic needle (#16), which was inserted into the ray opposite the host ray. In this way, the body wall of the host ray was left intact and the tissue was not likely to slip out immediately upon implantation. The cardiac stomach, and perhaps the pyloric as well, suffered some trauma when the needle passed through it, but its many folds and great elasticity served to occlude any openings through which the tissue might escape. With the needle in position, the trimmed donor caecum was drawn into the syringe with a small amount of sea water, the syringe was connected to the positioned needle and the tissue expelled into the coelom of the host. Care was taken not to introduce air bubbles into the coelom during this process. The advantage in first positioning the needle and then attaching the syringe lay primarily in the fact that there was less chance of macerating the tissue if it had to pass through the needle only once.

Two rays of an individual were generally used as host rays, and each was considered a separate experimental case. When an autologous transplant was being made, one of the rays opposite that intended to be the host ray was chosen as the donor ray, and the tissue was removed from it through a slit made in the aboral wall. The needle could later be inserted into the same incision. In some of the smaller specimens of *Asterias*, where it was difficult to slit the body wall accurately because of the small area of the aboral surface of the rays, the tip of the donor ray was cut off and the caecum removed through the open end. Thereafter, the procedure for the autologous transplants was identical to that for the others.

The hosts were examined daily for at least a week after implantation. Presence of edema, autotomy of host rays, and such signs of morbidity as shedding of patches of body wall or development of excessive flaccidity were noted. In addition, the dermal branchiae and oral and anal areas were checked for indications of the elimination of implanted tissue. At the end of a week (or longer in some cases), the hosts were dissected after relaxation in MgCl_2 , and the disposition of the trans-

plants noted. If the tissue was recovered, it was fixed in Bouin's solution for at least 24 hours (48, if it included calcareous elements), and after washing in 80% alcohol, was dehydrated, cleared, and embedded in paraffin. Sections were cut at 10 μ and stained with Mallory's phosphotungstic acid hematoxylin, which differentiates muscle, connective tissue, cell membranes, and secretion granules, and were examined for evidence of growth, necrosis, and other changes. Control pieces of caecum that had been passed through the needle without being implanted were similarly fixed, sectioned, and stained, in order to estimate the damage that might be caused to the tissue by the injection procedure.

RESULTS

a. Implantation experiments

When the animals were examined and dissected, it was found that the disposition of the donor tissue tended to fall into one of four categories, scored as follows:

(a) "Leaking"—The transplant caecum began to pile up in the dermal branchiae of the hosts, usually appearing here within one to three days after implantation. The distal halves of the branchiae, which had become swollen with donor tissue, often rounded up and pinched off from the bases, in effect undergoing autotomy. In other cases, the branchiae ruptured without autotomizing, so that the tissue simply passed out through the distal openings. In a few instances, where several adjacent branchiae had ruptured simultaneously, actual rifts in the body wall were formed, through which masses of implanted tissue could be seen emerging. In any case, when the hosts were dissected at the end of the week, the donor tissue was recovered, if at all, as a few small pieces projecting into the coelom from the bases of the branchiae.

(b) "Not recovered"—In many cases, the transplant was nowhere to be seen when the host was dissected. Parts of host caecum or body wall were stained with methylene blue, however, indicating that the donor tissue had been successfully injected into the coelom and had remained there long enough to impart some of its blue coloring to the surrounding host tissue. *Asterias* transplants tended to transfer more of the stain than *Patiria* transplants. It seems reasonable to assume that the tissue had in fact been eliminated, but by some route other than through the dermal branchiae.

(c) "Recovered"—The donor tissue was found apparently intact, either floating free in the coelom, or attached to the host body wall or pyloric caeca.

(d) "Lost"—When it was impossible to trace the disposition of the tissue, *i.e.*, when there was no recovery or evidence of leakage, and no staining of any host tissues could be noted, it was assumed that the tissue had not been successfully introduced into the coelom, and the data for that case were discarded. Into this category were also placed animals that had become moribund or had autotomized the host ray(s).

The first series of experiments involved transplanting pieces of homologous or heterologous pyloric caecum to determine the reaction of the hosts to transplants in general, and to see whether there were differences in the treatment of homologous and heterologous tissues. Table I presents a synopsis of the results of this series.

The figures for donor *Asterias* include both *A. forbesi* and *A. vulgaris*, between which no distinction was apparently made by either *Asterias* or *Patiria* hosts. These data indicate that both host species tend to tolerate their own tissue and to eliminate that of the other species. In order to confirm these observations, several double implantations were made, with each host receiving one homologous and one heterologous piece of caecum. Table II summarizes the results of these experiments. In the case of the single piece of *Henricia* caecum which was not recovered, the initial injection was probably unsuccessful.

TABLE I

Results of experiments demonstrating host reaction to single implants of pyloric caecum

Host	Donor	Leaking	Recovered	Not recovered	Total
<i>Patiria</i>	<i>Patiria</i>	3	29	0	32
	<i>Asterias</i>	0	1	13	14
	<i>Henricia</i>	9	1 (in frags.)	0	10
<i>Asterias</i>	<i>Asterias</i>	0	11	0	11
	<i>Henricia</i>	7	0	0	7

The results of both sets of experiments are mutually consistent and present evidence of what seems to be definite discrimination between homologous and heterologous transplants. These figures also demonstrate that the same animal can retain one transplant while eliminating another.

There are evident contrasts between the modes of elimination of *Asterias* and *Henricia* caecum by both *Patiria* and *Asterias*. *Patiria*, though it eliminates *Henricia* caecum through the branchiae, may dispose of *Asterias* tissue by pushing

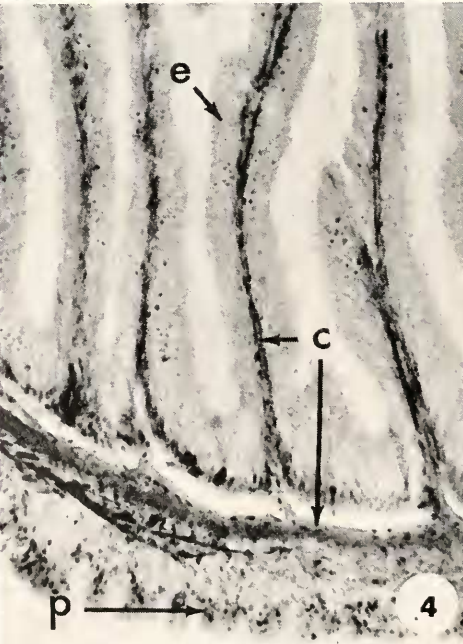
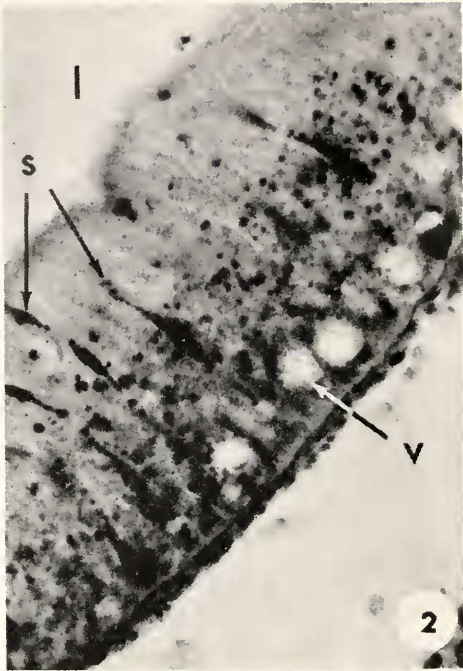
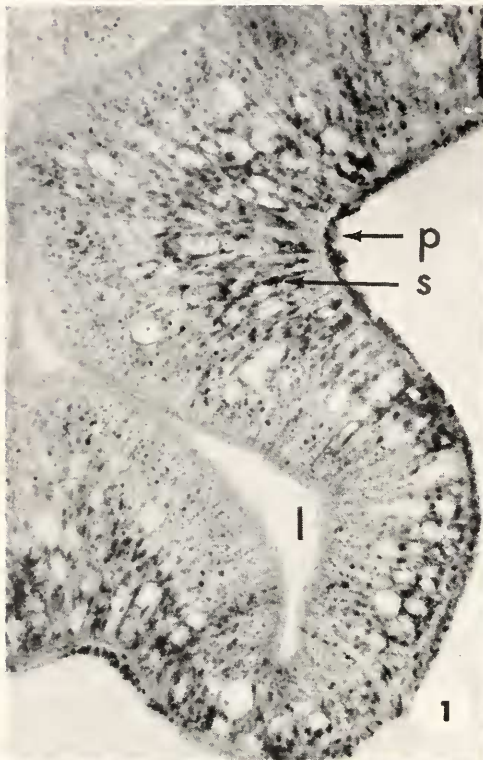
TABLE II

Results of experiments determining whether a single host discriminates between simultaneously implanted homologous and heterologous caecum

Host	Donor	Leaking	Recovered	Not recovered	Total
<i>Patiria</i>	<i>Patiria</i>	0	4	0	4
	<i>Asterias</i>	0	0	4	4
<i>Asterias</i>	<i>Asterias</i>	0	11	0	11
	<i>Henricia</i>	9	1	1	11

it back into the cardiac stomach, for on several occasions, transplanted tissue was recovered projecting from the mouth. *Asterias* also eliminates *Henricia* caecum through the branchiae. Unfortunately, transportation difficulties made it impossible to obtain an adequate supply of *Asterias* in Ithaca, and I was unable to determine how this species reacts to *Patiria* implants.

To determine whether tolerance of homologous material in the coelom extended to tissue taken from other parts of the body, observations were made on homologous transplants of tissue other than pyloric caecum. Eight *Patiria* host rays were im-



FIGURES 1-4.

planted with homologous rectal caecum, an organ normally found in the aboral region of the central disc. All eight transplants were recovered in apparently healthy condition, and in one case, the implanted rectal caecum was joined to an adjacent pyloric caecum of the host.

On other occasions, recovered pyloric caecum transplants, especially older ones which had spent three or more weeks in the host coelom, had fragments of ossicles embedded within them. The sources of these fragments are uncertain; perhaps they had been picked up and carried in by the tip of the needle during the implantation procedure. This retention of both rectal caecum and ossicle fragments, despite their considerable physical differences from pyloric caecum, supports the view that elimination cannot be based on physical differences alone. It also suggests that the animal is not sensitive to the presence of tissue elements in the wrong places in the body.

b. Histological observations

Normal pyloric caecum is composed of an outer peritoneum of cuboidal cells, layers of muscular, connective and nervous tissue, and an inner epithelium of tall flagellated columnar cells, this layer containing mucous gland cells and secretory cells producing vesicles and granules (Anderson, 1953; see also Fig. 1). Examination of pieces of control caecum which had been passed through the needle before fixation revealed areas of tissue normal in every respect, interspersed with places where the tissue had been broken and abraded by the injection process.

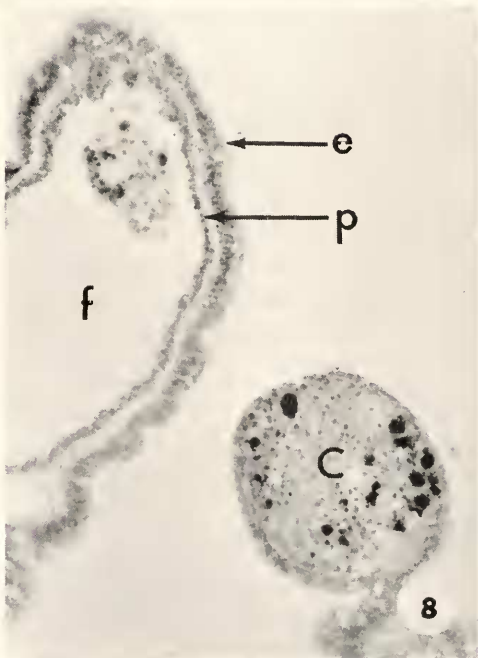
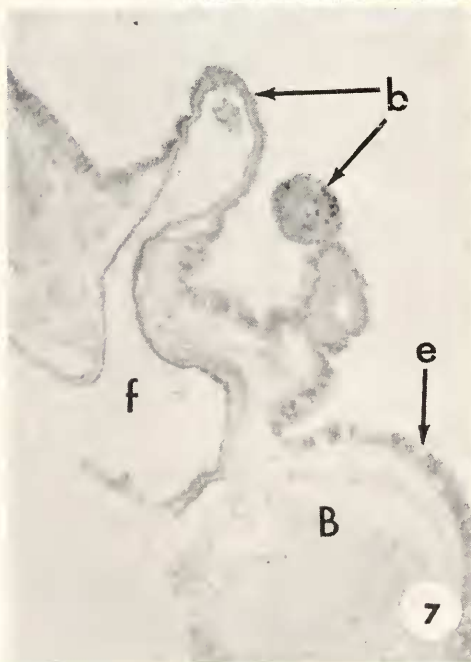
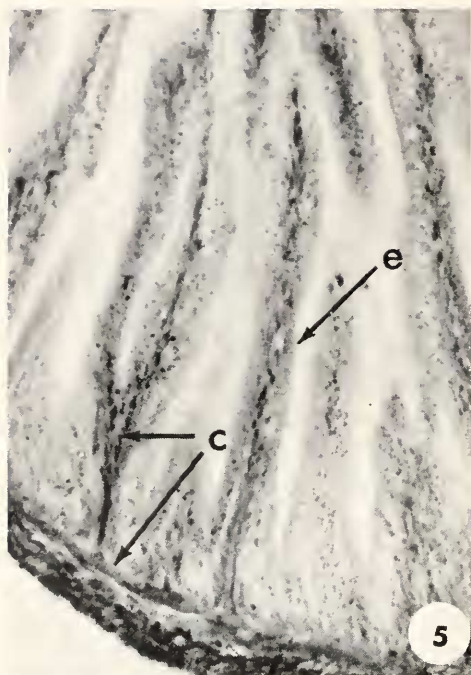
Both *Patiria* and *Asterias* pyloric caecum transplants recovered after one to five weeks in homologous hosts generally showed the same mixed appearance as the injected and fixed controls, healthy areas alternating with damaged ones. The sound areas (Fig. 2) were generally indistinguishable from normal tissue. The presence of prominent secretion granules in the recovered caecum (Fig. 2) indicates that metabolic activities were continuing at the time of fixation. There were few amoebocytes associated with the intact areas. Damaged areas, on the other hand, showed changes, particularly evidence of necrosis and disorganization associated with amoebocytes. The amoebocytes were often rounded and filled with what appeared to be cellular debris, including recognizable secretion granules. Whether these phagocytes came from the donor, the host, or both could not be determined.

FIGURE 1. *Patiria*, normal pyloric caecum. Many of the tall columnar epithelial cells contain secretion granules and vesicles. $\times 260$. l, lumen of caecum; p, peritoneum; s, secretory cell with granules.

FIGURE 2. *Patiria*, pyloric caecum recovered after four weeks in a homologous host. There is no evidence of damage or necrosis; the secretory cells show evidence of continuing normal metabolic activity. $\times 520$. l, lumen of caecum; s, secretory cell with granules; v, vesicle.

FIGURE 3. *Patiria*, pyloric caecum with attached mass of amoebocytes and connective tissue recovered from a homologous host after four weeks. The mass of new cells is associated with damaged areas of caecum (not shown) but has extended to join the healthy caecum at left. The connective tissue fibers in the new tissue are continuous with those in the donor caecum. Peritoneum, also continuous with that of the donor caecum, covers the amoebocyte mass. $\times 260$. a, amoebocyte mass; C, donor caecum; c, connective tissue fibers; l, lumen of caecum; p, peritoneum.

FIGURE 4. *Patiria*, normal rectal caecum. The inner epithelium is thrown into folds supported by connective tissue partitions. $\times 260$. c, connective tissue fibers; e, epithelium; p, peritoneum.



FIGURES 5-8.

Recovered transplants were found joined to the host caecum or body wall on several occasions. In some specimens, connections were made at several points. The more tenuous of these links appeared to be established by amoebocytes alone, while more substantial bridges showed connective tissue fibers in addition. It seems probable that the amoebocytes established the first bridges, with connective tissue fibers invading later.

Some of the older transplants (four or five weeks) had masses of amoebocytes and connective tissue filling in the gaps or folds in the implant (Fig. 3). This growth was often quite extensive and gave the impression of rounding out the contours of the transplant. In some cases, peritoneal epithelium could be seen around the mass of new cells (Fig. 3). The development of the amoebocyte masses usually began on the borders of the damaged sections, occasionally extending to the healthy areas as well. Where ossicles were embedded in the transplant, they were also associated with amoebocytes and connective tissue, and appeared to be firmly joined to the surrounding caecum. In no case, however, did the recovered homologous tissue give the impression of undergoing encapsulation, for areas not obviously damaged were often free of amoebocyte activity.

Normal rectal caecum shows the same layers as normal pyloric caecum, but the inner epithelium is greatly folded, each fold being supported by inward extensions of the connective tissue layer (Fig. 4). Rectal caecum recovered after a week (Fig. 5) was for the most part indistinguishable from the normal caecum, although it did show some local damage associated with amoebocyte activity and connective tissue formation similar to that described in the recovered pyloric caecum.

One case of encapsulation was observed, involving a small fragment of *Asterias* transplant recovered from a host *Patiria*. Sections showed a small core of extremely disorganized donor tissue with a few scattered secretion granules remaining, surrounded by amoebocytes. There was no evidence of connective tissue formation.

Examination of sections of body wall in which the dermal branchiae were eliminating *Henricia* caecum showed interesting details. The branchiae were rounded and swollen at their tips, which either ruptured (Fig. 6), or pinched off (Figs. 7, 8). The walls of the swollen parts were stretched quite thin compared with the more typical branchiae (Fig. 8); the columnar epidermis appeared almost cuboidal, the connective tissue layer was greatly attenuated, and the peritoneum was vague and indistinct. It was impossible to distinguish the muscle layers at all.

FIGURE 5. *Patiria*, rectal caecum recovered from homologous host after one week (slightly oblique section). This tissue is virtually indistinguishable from the normal rectal caecum (Fig. 4). $\times 260$. c, connective tissue fibers; e, epithelium.

FIGURE 6. *Henricia* caecum being eliminated through ruptured *Asterias* branchia. (The section is not quite parallel to the long axis of the branchia.) This branchia and the ones on either side are packed with *Henricia* caecum. Note the expansion of the tissue already out of the branchia. $\times 90$. B, body wall; b, branchial wall; C, *Henricia* caecum.

FIGURE 7. *Asterias* branchia eliminating *Henricia* caecum. (Section is slightly tangential to the long axis of the branchia.) The tip of the branchia containing the *Henricia* caecum is separated from the rest by a prominent constriction. Note the wrinkling of the base of the branchia. $\times 90$. B, body wall; b, branchiae; e, epidermis; f, coelom.

FIGURE 8. Detail of Figure 7. Note how the wall of the autotomizing portion of the branchia has been stretched. (Compare with the more typical branchia at left.) $\times 260$. C, *Henricia* caecum; e, epidermis of branchia; f, coelom (cavity of branchia); p, peritoneum.

In contrast, the bases of the branchiae, below the point of constriction, appeared quite normal histologically, except that they were slightly wrinkled transversely (Fig. 7) as if either circular or longitudinal muscles or both were somewhat contracted. Where the branchiae had ruptured, *Henricia* caecum could be seen squeezing through the channel provided by the branchial walls and escaping to the outside (Fig. 6). The caecum in the branchiae was not in fragments, but rather gave the impression of being folded and packed into the lumen of the organ (Fig. 6). None of the *Henricia* caecum appeared to have unusual numbers of amebocytes associated with it, although it was clearly being actively eliminated.

DISCUSSION

The results of these experiments suggest that there are at least three different means of elimination of foreign materials from the asteroid coelom. The first, amebocytic attack, was expected from accounts in the literature. The second, elimination of tissue through rupturing or autotomizing branchiae, was somewhat of a surprise, for although Lison (1930) reported branchial autotomy in response to the presence of large clumps of amebocytes in the branchiae, I am aware of no other report of branchial elimination of pieces of tissue. The third method, that still undetermined means by which *Patiria* eliminated *Asterias* pyloric caecum (possibly through the cardiac stomach, although I have no histological evidence that this was the case), was also not predictable on the basis of previous reports of elimination of material.

Phagocytosis of particulate matter may be demonstrated easily in both *Patiria* and *Asterias* by injection of a carmine suspension into the coelom; the branchiae shortly become filled with amebocytes bearing ingested carmine particles. However, amebocytic attack and invasion cannot be responsible for the branchial elimination of *Henricia* caecum, for the tissue in the branchiae remains in a relatively intact state and has few amebocytes associated with it. Nor does encapsulation by amebocytes seem to be a common response to the presence of large foreign bodies in the coelom, for it was observed only once during the whole study.

It was not possible to determine at this time just why *Henricia* caecum was selectively eliminated through the branchiae by both hosts. The mechanisms involved might depend upon physical or chemical differences, or both, between this caecum and those of *Asterias* and *Patiria*. In any case, this means of elimination seems to be fairly important to both hosts; at least it was consistently employed.

The filling and rupturing of the branchiae must require a considerable amount of force, perhaps more than can be accounted for on the basis of ciliary currents alone. The musculature of the branchiae, consisting of both longitudinal and circular fibers, might be aiding the movement of the tissue to the tip, perhaps by peristaltic action. The longitudinal muscles normally retract the branchiae in response to external disturbances, and the circular muscles are antagonistic to the longitudinal muscles. However, the fact that an autotomizing branchia may be pinching in strongly at one level alone and nowhere else along its length implies rather specific control over local contraction, which in turn suggests that peristaltic movement is at least theoretically possible. The wrinkled appearance of the autotomizing branchiae below the point of constriction, as seen in Figure 7, might indicate that such contraction was going on at the time the tissue was fixed.

In addition to eliminating material through the branchiae, *Asterias* can quickly and easily autotomize whole rays. *Patiria* cannot autotomize easily, but the efficiency with which it evidently eliminated the *Asterias* transplants suggests that it is well protected despite its inability to shed rays. To determine whether *Asterias* can also eliminate material through the cardiac stomach (if this is in fact how *Patiria* eliminates the *Asterias* tissue) or whether this species relies entirely on use of the branchiae and on autotomy requires further experimentation.

The question of the level (*i.e.*, tissue, organ, or higher) at which these elimination mechanisms are operating is experimentally beyond the scope of the present study, but it presents possibilities for future investigations. Elimination through the branchiae, for example, may be a complex process; it remains to be determined whether it is an automatic local reaction to the presence of anything in the branchiae, or whether the process is selective and under the regulation of the animal as a whole. If the latter is the case, one might well consider it an integrated response. It is also important to remember that local and general effects may be occurring simultaneously and that demonstration of the one does not preclude the existence of the other.

The role of the amebocytes associated with the damaged areas of the recovered caecum may be a multiple one. It is unlikely that they are encapsulating the implant, walling it off from the coelom by forming a shell around it, as this process was found only once in this series of experiments. They are almost certainly engaged in cleaning up cellular debris in these damaged areas. They might also be participating in regeneration of fragmented caecum. Anderson (1962), studying the regeneration of pyloric caeca in *Henricia leviuscula*, discusses the possibility that the amebocytes might actually be differentiating and supplying cells for the new caecum. Here, also, more work must be done to understand what is taking place.

The results of the present preliminary study may or may not indicate that responses of an immunological nature are evoked by tissue implants. The response of the host does seem to vary in relation to the source of the implanted tissue. No distinction was made (within the framework of present experiments) between autologous and homologous tissue, or between tissue of two closely related species, *Asterias forbesi* and *Asterias vulgaris*. On the other hand, there was discrimination between members of two different families in the same order (*Henricia* and *Patiria*), and between members of two different orders (*Asterias* and the other two). These latter results are in agreement with those of Anderson (unpublished) on the rejection of *Henricia* caecum by *Patiria*. They are contrary, however, to his findings on the *Pisaster* implants, which were evidently tolerated by *Patiria*. This discrepancy is unexplained and might conceivably be another example of the "disconcerting inconsistency" mentioned by Cantacuzène (1923) as characteristic of invertebrate reactions.

The mechanism of distinction is obscure, but as metabolizing caecum is undoubtedly exchanging molecules with the coelomic fluid (Ferguson, 1964a, 1964b), the host may be reacting to the presence of atypical molecules in the coelomic fluid. On this basis, also, the distinction between "tissue" and "general" levels of reaction becomes a pertinent question.

Much more work remains to confirm and extend these results. No attempt

was made to follow the progress of any of the transplants for longer than five weeks. It may be that all foreign tissue is eventually rejected or perhaps encapsulated if enough time is provided, although the experimental data suggest that this is probably not the case. Other donor-host combinations should be tried to determine how specific and how consistent these reactions actually are and where the lines are drawn between tolerance and rejection. The role of the amebocytes needs clarification with respect to such questions as whether they are only removing debris, or are also taking part in regeneration of tissue in the damaged areas of caecum. Until these questions are answered, the precise bases of these reactions toward foreign implants will remain unknown.

SUMMARY

1. Two starfishes, *Patiria miniata* and *Asterias forbesi*, were found to discriminate between coelomic implants of homologous and heterologous pyloric caecum obtained from donors of these species and of two others, *Asterias vulgaris* and *Henricia sanguinolenta*.

2. Homologous transplants were recovered from the hosts one to five weeks after implantation and were found to be generally normal in histological appearance, although there was some growth of connective tissue and amebocyte masses in areas which had probably been damaged during the implantation procedure. Heterologous transplants were eliminated by the hosts within a week of implantation.

3. *Henricia* caecum was eliminated through the dermal branchiae by both hosts. *Patiria*, in contrast, eliminated *Asterias* caecum in a still undetermined way; the tissue may have been transferred from the host ray to the cardiac stomach, and either digested there or passed out through the mouth.

4. Contrary to expectation, amebocytic attack, phagocytosis, and encapsulation were not seen to play an important role in elimination of heterologous transplants, although amebocytes were associated with damaged areas in the homologous transplants. Such amebocytic activity is probably concerned with removal of cellular debris.

5. The results of these experiments may express a general tendency of starfishes to distinguish between implants of donors of different degrees of relationship, or they may represent special cases of discrimination. More work remains to determine how specific these tolerance-rejection thresholds are and how consistently they vary with different host-donor combinations.

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